

Amendments

In the Claims:

Claims 1-45 (Canceled).

46 (Currently amended): An isolated, linear nucleic acid molecule comprising, in order: (a) at least one first topoisomerase recognition site; (b) at least one first recombination site; (c) at least one nucleic acid segment; (d) at least one second recombination site; and (e) at least one second topoisomerase recognition site, wherein said first topoisomerase recognition site and said first recombination site are separated from each other by about 0 to about 100 nucleotides.

47 (Canceled).

48 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are the same.

49 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are different.

50 (Previously presented): The nucleic acid molecule of claim 46, wherein the separation between said first topoisomerase recognition site and said first recombination site is selected from the group consisting of about 0 to about 50 nucleotides; about 0 to about 30 nucleotides; about 0 to about 20 nucleotides; and about 0 to about 10 nucleotides.

51 (Previously presented): The nucleic acid molecule of claim 46, wherein the separation between said first topoisomerase recognition site and said first recombination site is about 0 to about 50 nucleotides.

52 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are selected from the group consisting of:

- (a) *attB* sites;
- (b) *attP* sites;
- (c) *attL* sites;
- (d) *attR* sites;
- (e) *lox* sites;
- (f) *psi* sites;
- (g) *dif* sites;
- (h) *cer* sites; and
- (i) *frt* sites.

53 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are *lox* sites.

54 (Previously presented): The nucleic acid molecule of claim 46, wherein said at least one nucleic acid segment comprises at least one open reading frame.

55 (Previously presented): The nucleic acid molecule of claim 54, wherein said open reading frame encodes an antibiotic resistance gene.

56 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and/or said second topoisomerase recognition sites are recognized and bound by a type I topoisomerase.

57 (Previously presented): The nucleic acid molecule of claim 56, wherein said type I topoisomerase is a type IB topoisomerase.

58 (Previously presented): The nucleic acid molecule of claim 57, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.

59 (Previously presented): The nucleic acid molecule of claim 58, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.

60 (Previously presented): A vector comprising the nucleic acid molecule of claim 46.

61 (Previously presented): The vector of claim 60, wherein said vector is an expression vector.

62-64 (Canceled).

65 (Currently amended): An isolated, linear nucleic acid molecule comprising:

- (a) a first recombination site and a second recombination site; and
- (b) a topoisomerase recognition site,

wherein the first or second recombination site is separated from the topoisomerase recognition site by about 0 to about 100 nucleotides.

66 (Canceled).

67 (Previously presented): The nucleic acid molecule of claim 65, wherein said first or second recombination site is separated from said topoisomerase recognition site by about 0 to about 10 nucleotides.

68 (Previously presented): The nucleic acid molecule of claim 65, wherein said first or second recombination site is separated from said topoisomerase recognition site by about 10 to about 30 nucleotides.

69 (Previously presented): The nucleic acid molecule of claim 65, wherein said first or second recombination site is separated from said topoisomerase recognition site by about 20 to about 50 nucleotides.

70 (Previously presented): The nucleic acid molecule of claim 65, wherein said first or second recombination site is separated from said topoisomerase recognition site by about 40 to about 80 nucleotides.

71 (Previously presented): The nucleic acid molecule of claim 65, wherein said first or second recombination site is separated from said topoisomerase recognition site by about 70 to about 100 nucleotides.

72 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are selected from the group consisting of:

- (a) an *attB* site;
- (b) an *attP* site;
- (c) an *attL* site;
- (d) an *attR* site;
- (e) a *lox* site;
- (f) a *psi* site;
- (g) a *dif* site;
- (h) a *cer* site; and
- (i) an *frt* site.

73 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are *lox* sites.

74 (Previously presented): The nucleic acid molecule of claim 65, wherein said topoisomerase recognition site is recognized and bound by a type I topoisomerase.

75 (Previously presented): The nucleic acid molecule of claim 74, wherein said type I topoisomerase is a type IB topoisomerase.

76 (Previously presented): The nucleic acid molecule of claim 75, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.

77 (Previously presented): The nucleic acid molecule of claim 76, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.

78 (Previously presented): A vector comprising the nucleic acid molecule of claim 65.

79 (Previously presented): The vector of claim 78, wherein said vector is an expression vector.

80 (Previously presented): A host cell comprising the isolated nucleic acid molecule of claim 65.

81 (Previously presented): A host cell comprising the vector of claim 78.

82 (Previously presented): A host cell comprising the vector of claim 79.

83 (Previously presented): A kit comprising the isolated nucleic acid molecule of claim 46.

84 (Previously presented): A kit comprising the isolated nucleic acid molecule of claim 65.

85 (Previously presented): The kit of claim 83 or claim 84, further comprising one or more components selected from the group consisting of one or more topoisomerases, one or more recombination proteins, one or more vectors, one or more polypeptides having polymerase activity, and one or more host cells.

86 (Previously presented): A vector selected from the group consisting of pcDNAGW-DT(sc), pENTR-DT(sc), pcDNA-DEST41, pENTR/D-TOPO, pENTR/SD/D-TOPO, pcDNA3.2/V5/GWD-TOPO and pcDNA6.2/V5/GWD-TOPO.

87 (Previously presented): A host cell comprising the vector of claim 86.

88 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are *att* sites.

89 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are mutants, variants, and derivatives of *att* sites.

90 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are *loxP* or *loxP511* sites.

91 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are selected from the group consisting of *attB* sites, *attP* sites, *attL* sites and *attR* sites.

92 (Previously presented): The nucleic acid molecule of claim 46, wherein said first and second recombination sites are mutants, variants, and derivatives of *attB* sites, mutants, variants, and derivatives of *attP* sites, mutants, variants, and derivatives of *attL* sites, or mutants, variants, and derivatives of *attR* sites.

93 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are *att* sites.

94 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are mutants, variants, and derivatives of *att* sites.

95 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are *loxP* or *loxP511* sites.

96 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are selected from the group consisting of *attB* sites, *attP* sites, *attL* sites and *attR* sites.

97 (Previously presented): The nucleic acid molecule of claim 65, wherein said first and second recombination sites are mutants, variants, and derivatives of *attB* sites, mutants, variants, and derivatives of *attP* sites, mutants, variants, and derivatives of *attL* sites, or mutants, variants, and derivatives of *attR* sites.

98 (New): An isolated nucleic acid molecule comprising, in order: (a) at least one first topoisomerase recognition site; (b) at least one first recombination site; (c) at least one nucleic acid segment; (d) at least one second recombination site; and (e) at least one second topoisomerase recognition site, wherein said first topoisomerase recognition site and said

first recombination site are separated from each other by about 0 to about 100 nucleotides and wherein said first and second recombination sites are different.

99 (New): The nucleic acid molecule of claim 98, wherein said first and second recombination sites are selected from the group consisting of:

- (a) *attB* sites;
- (b) *attP* sites;
- (c) *attL* sites;
- (d) *attR* sites;
- (e) *lox* sites;
- (f) *psi* sites;
- (g) *dif* sites;
- (h) *cer* sites; and
- (i) *frt* sites.

100 (New): The nucleic acid molecule of claim 98, wherein said first and/or said second topoisomerase recognition sites are recognized and bound by a type I topoisomerase.

101 (New): The nucleic acid molecule of claim 100, wherein said type I topoisomerase is a type IB topoisomerase.

102 (New): The nucleic acid molecule of claim 101, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.

103 (New): The nucleic acid molecule of claim 102, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.

104 (New): A host cell comprising the isolated nucleic acid molecule of claim 98.

105 (New): A host cell comprising a vector, the vector further comprising the nucleic acid molecule of claim 98.

106 (New): The host cell of claim 105 wherein the vector is an expression vector.